ELECTRICAL POLARITY AND AUXIN TRANSPORT

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(WITH TEN FIGURES)

Introduction

The polar basal transport of the growth substances (auxins, growth hormones) in plants is a well known phenomenon, demonstrated first by WENT (**81**), and studied in detail by VAN DER WEIJ (**78, 80**). These investigators used the *Avena* (oat) coleoptile.[1] That the phenomenon is more or less general is indicated by the polar transport of auxin in roots (CHOLODNY, **19**; NAGAO **60**); in hypocotyls of *Raphanus* (VAN OVERBEEK, **62**), of *Pisum* (SKOOG, **74**); in leaves (AVERY, **2**); in the coleoptile of *Avena* (WENT, **81**; LAIBACH and KORNMANN, **40**; VAN DER WEIJ, **78, 80**; and SKOOG, **74**) and corn (VAN OVERBEEK, **63**); in *Elaeagnus* (woody cutting) (VAN DER WEIJ, **79**); in stems of *Coleus, Vicia,* and *Phaseolus;* and in hypocotyls of *Vicia, Phaseolus,* and *Lupinus* (MAI, **54**).

Other workers have reported non-polar transport of auxin in plants. HITCHCOCK and ZIMMERMAN (**31**) and ZIMMERMAN and WILCOXON (**85**) have shown an apical transport of heteroauxin (indole-3-acetic acid) and several other active compounds in stems of *Helianthus tuberosus, Nicotiana tabacum,* and in *Lycopersicum esculentum,* as indicated by induction of adventitious roots and by epinastic response of leaves. LOEHWING and BAUGUESS (**45**) have shown that heteroauxin could be absorbed by the root system of potted seedlings of *Matthiola incana* and be transported apically to increase the stem elongation over that of the controls. Both the Boyce-Thompson workers and LOEHWING and BAUGUESS have merely shown that auxin applied in high concentrations can be carried in the transpiration stream. This, of course, will not give polar transport. Higher concentrations of auxin may have effects which are not normally encountered. For example, high concentrations applied at the base of *Pisum* cuttings induce roots there, whereas in the lower, more physiological concentrations roots may be induced at the bases, only by applying auxin at the morphological tips (WENT and THIMANN, **83**).

LAIBACH and FISCHNICH (**41**) have shown that the transport of heteroauxin was not strictly polar in leaves of *Coleus* and in cotyledons of *Cucumis sativa,* but that transport could occur apically. The apical transport was much smaller than basal transport, however. AVERY (**2**), as mentioned above, found only basal transport. AVERY determined this by diffusion of

[1] Coleoptiles are leaf-sheaths which envelop growing points and first foliage leaves of grass seedlings.

the auxin occurring naturally in the leaves, whereas LAIBACH and FISCHNICH applied heteroauxin in a concentration of 0.5 per cent. in lanolin, an altogether unphysiological concentration.

JOST and REISZ (**34**) demonstrated apical transport of high concentrations of heteroauxin in *Avena* coleoptiles. This was seen from the growth of sections with their basal ends immersed in the auxin solutions, and from actual transport experiments in which auxin was collected in agar blocks at the apical ends of sections supplied with auxin in agar at the basal ends. The apical transport was, however, much less pronounced than the normal basal transport, and hence semipolarity still exists. The concentrations of auxin used in the transport experiments was 1:200,000, the length of the sections 10 mm., and the time of transport overnight. As to its effect on growth, the apical transport may have been effected by capillarity in the hollow coleoptiles. Regardless of these findings, WENT, VAN DER WEIJ, and others always observed strictly basal transport in short sections (1 to 4 or 5 mm.) when shorter periods of time (1 to 3 hrs.) were used. Moreover, as VAN DER WEIJ (**78**) mentioned, when higher concentrations of auxin are used (1:200,000), the auxin may be transported by capillarity in films of water condensed on the surfaces of the sections. At any rate, although strict polarity is always difficult to observe when high concentrations of auxin are used, the polarity still dominates apical transport. The concentrations ordinarily used are of the order of $1:10^6$ or less (WENT and THIMANN, **83**).

As has been mentioned, CHOLODNY (**19**) and NAGAO (**60**) demonstrated a polar basal transport in roots. GORTER (**28**), DE HAAN (**30**), and others claim otherwise. These controversial statements will not be discussed at this point, since they have no bearing on the polar basal transport in *Avena*.

The mechanism of polar transport

The mechanism of this polar transport is as yet little understood. WENT (**81**) showed that the transport was always in a basal direction in the *Avena* coleoptile when physiological concentrations were used; that in his special case, the velocity of this transport was about two hundred times greater than that of ordinary diffusion (being 10 mm. per hour); and that its polarity was unaffected by gravity. VAN DER WEIJ (**78, 80**) confirmed these findings and, in addition, showed that the transport would occur against a considerable concentration gradient, suffering no appreciable change. He also found that the velocity of the transport was reversibly lowered to that of diffusion when the temperature was lowered to 0° C. At this temperature, however, the polarity of the transport persisted. Polarity, on the other hand, was reversibly abolished by ether narcosis (VAN DER WEIJ, **80**). BONNER (**6, 7**) indicated that transport was dependent

upon the presence of oxygen. With regard to the independence of transport and gravity, PFAELTZER (**64**) found that 14.5 × gravity, produced by a centrifugal field, had no effect on polar transport in the *Avena* coleoptile.

PROTOPLASMIC STREAMING AS THE MECHANISM

VAN DER WEIJ (**70**) concluded that polar transport was a "vital process," but that protoplasmic streaming had nothing to do with it, since the velocity of transport was independent of temperature down to very low values, *e.g.* 0° C., while the velocity of protoplasmic streaming depended upon temperature within wide limits, citing LAMBERS (**42**). This view is supported by the observation of SCHUMACHER (**73**) that fluorescein shows polar diffusion in the plasma of stem hairs of *Cucurbita pepo,* the rate and direction of this transport being constant, while the rate and direction of protoplasmic streaming varied. BOTTELIER (**9**) favored some correlation between protoplasmic streaming and transport, finding the *velocity* of streaming (3 cm. per hour) to be *constant* between 17° and 35° C., while the amount of protoplasm in actual rotation (streaming intensity) increased with temperature, just as transport intensity increases (VAN DER WEIJ, **78**). Furthermore the velocity of transport (1 cm. per hour) is too great to be explained by a diffusion process unless it is of the nature of the model described by VAN DEN HONERT (*cf.* below). BOTTELIER (**10**) also found that oxygen limited protoplasmic streaming, as it presumably does transport.

From this discussion it is probable that protoplasmic streaming has nothing to do with the *polarity* of transport, but may be a factor in its *velocity.*

ACTIVATED DIFFUSION AS THE MECHANISM

A possible mechanism for transport is that suggested by several workers (*cf.* BRINKMAN and SZENT-GYÖRGYI, **16**; VAN DEN HONERT, **32**; SÖLLNER, **75**), which demonstrates the transport of surface-active substances at interfaces whose interfacial tension has been lowered at one end by the addition of these substances ("spreading"). The velocity of the transport of KOH in VAN DEN HONERT'S model was in one case 68,000 times greater than that of ordinary diffusion of KOH.

MASON and MASKELL (**57**) found that the diffusion of sugar in the cortex of the cotton plant complies with the rules of concentration gradients and directional flow for diffusion, but that the velocity was between 20,000 and 40,000 times greater than the ordinary diffusion of sugar would be expected to exhibit. MASON and PHILLIS (**58**) found that oxygen was necessary for such transport in the cotton plant, and state: "It is suggested that the mechanism activating diffusion consists in some special organization in the cytoplasm, maintained by metabolic energy, whereby the resistance to solute movement is so reduced that materials diffuse in the sieve-tube at rates com-

parable with those in a gas." PHILLIS and MASON (**66**), moreover, have shown that sucrose is transported against a concentration gradient in the leaf of the cotton plant. This recalls the similar transport of auxin against a concentration gradient in the *Avena* coleoptile, but is different in that the sugar transport is not as polar. The "organization" spoken of by MASON and PHILLIS is as yet unknown, although the model of VAN DEN HONERT is suggestive.

Hence "activated diffusion" may be a factor in determining the velocity of auxin transport in plants, particularly since auxin is surface active (OKUNUKI, **61**; KÖGL, ERXLEBEN, and HAAGEN-SMIT, **39**), but it is difficult to see how such a mechanism could explain polarity.

Electrical polarity

BRAUNER (**13**) found that the underside of horizontally placed plants became electropositive to the upper side. He also found that the shaded side of illuminated seedlings developed an electropositivity with respect to the illuminated side. Later BRAUNER and BÜNNING (**15**) correlated the geo-electric effect with electrotropisms. CHOLODNY (**18**) had already developed the theory that the plant growth hormone is electrically transported in the plant, accumulating more on one side than on the other, thus causing differential growth and a tropism. DOLK (**22**) assumed that the growth hormone was an acid, and suggested that the dissociated anion would be transported to the geo- or photo-induced positive pole. For a review of this literature, see WENT (**82**).

From a survey of older literature on the subject of electrical polarity in living organisms, WENT (**82**) formulated in his "Botanische Polaritätstheorie" the idea that the dissociated anion of auxin is transported longitudinally in the plant as a result of the inherent electrical polarity of the organ in question. By this time it was known that auxin was a weak acid (KÖGL and HAAGEN-SMIT, **38**). Applying his theory to seedlings, WENT suggested that the apical end of a seedling was electronegative to the basal end, and that auxin anions were electrically and polarly transported basalward. He supported this theory with experiments demonstrating that *Impatiens* cuttings exhibited a bipolar staining in acid and basic dyes (*cf.* experimental part of this paper).

Previous indirect evidence has seemed to favor a causal relation between lateral transport of auxin and bioelectric potentials in plants (BRAUNER and BÜNNING, **15**; AMLONG, **1**; KOCH, **37**; DE HAAN, **30**).

For a general discussion of transport and of the polarity theories, the reader is referred to BOYSEN-JENSEN (**12**) and WENT and THIMANN (**83**).

In conclusion, it may be said that evidence seems to favor both protoplasmic streaming and activated diffusion as velocity components in trans-

port, and bioelectric potentials as the cause of polarity in the transport of auxin in plants.

It is the purpose of the present papers to reexamine the possibility of a linkage between electrical polarity and the polar transport of auxin in plants, the first paper being concerned with electrical polarity, and the second (see July, 1937, issue) with transport and electrical polarity.

A. Inherent electrical polarity

1. Polar dye uptake.—As mentioned in the introduction, Went (**81**), to substantiate his electrical polarity theory, demonstrated a bi-polar uptake of dyes in *Impatiens* cuttings. Negatively charged (acid) dyes penetrated most at the cut apices, and positively charged (basic) dyes at the bases of immersed cuttings. Referring to his paper, it is seen that his acid dyes included light-green, acid green, quinolin yellow, and methyl orange; while the basic dyes included safranin, methyl violet, prune pure, neutral red, thionin, and gentian violet. De Haan (**30**) investigated differential staining in geotropically bending *Vicia* roots and found that basic dyes accumulated most on the convex side, as would be expected from Brauner's (**13**) finding that this side was electropositive to the concave side. De Haan classified light-green and methyl orange as "anode-coloring" (basic) which contradicts Went's classification.

With respect to such controversies, electrophoretic experiments were performed with all of the dyes used in the experiments about to be described. The dye solutions (0.5 per cent.) were made up in distilled water at pH 6.0, and a current passed from zinc electrodes through U-tubes containing the dyes. The products of electrolysis at the electrodes were washed away by automatic siphons during the current passage. At the end of the experiments, the pH values at the electrodes were found to be unchanged. When such experiments were performed, all dyes classified as acids were found to be cathodic and all basic dyes anodic. Keller has said, however, that dye particles reverse their charges in protoplasm due to the fact that colloids adsorb the particles and impart to them the charge of the colloidal particle (Keller, **35, 36**; Gicklhorn and Keller, **27**). An exception exists, he says, when the dye is in such excess that the charge of the adsorbed dye particles neutralizes that of the colloidal particle. In this case the charge of the dye particle is not reversed. Lauer (**43**) could not confirm Keller's claim that protoplasm reverses the dye-particle charge. De Haan himself found a lack of agreement between Keller's tables and "test-object" *Hedera helix*, used by Keller in making up these tables. Owing to such uncertainties, it was assumed that the present electrophoretic experiments, conducted at pH 6.0 (which is approximately that of the cell content in plants), gave the true charge of the dyes used in the following experiments with plant cut-

tings. It was likewise assumed that the plant did not alter the sign of the charge on the particles. Electrical measurements later bore out this last assumption.

PRINGSHEIM (67) criticized WENT's dye-uptake experiments in that the distances penetrated (1 mm. at best) observed by WENT could not allow conclusions to be drawn. According to this criticism, DE HAAN's observations, based on the number of cells stained in cross sections of *Vicia* roots, would be even less valid. From such considerations, it became of interest to repeat WENT's experiments and to test these findings with electrical measurements. The following descriptions show that WENT's observations can be clearly duplicated.

TABLE I

BIPOLAR DYE UPTAKE IN *IMPATIENS* CUTTINGS

KEY:

0 ..... No staining
2 ..... Slight staining
4 ..... Up to 0.5 mm. penetration
6 ..... 1 mm. penetration
8 ..... 1.5 mm. "
10 ..... 2.0 mm. "
14 ..... 3.0 mm. "

DYE	PENETRATION	
	APEX	BASE
Negative dyes		
Trypan blue	2.0	3.0
Light-green	7.0	0.5
Methyl blue	5.0	2.0
Methyl orange	7.0	2.5
Congo red (colloidal)	4.5	6.5
Orange-G	10.0	1.0
Average	6.0	2.6
Positive dyes		
Safranin	7.0	9.3
Methyl violet	5.0	11.0
Neutral red	3.5	11.0
Janus green	5.0	9.0
Bismark brown	4.0	9.0
Thionin	5.0	7.0
Methylene blue	3.5	12.5
Cresyl violet	4.0	8.5
Brilliant cresyl blue	6.0	11.0
Nile blue A	8.0	14.0
Average	5.1	10.2

Etiolated *Impatiens balsamina* seedlings grown in sand in the dark room at constant temperature and humidity, were prepared by cutting away the cotyledons and the parts underground. Two to three cut hypocotyls were placed in upright test tubes filled with the dye solutions in 1 per cent. sucrose. The experiments were run in the dark 15 to 24 hours. After this time the hypocotyls were removed and examined in daylight, the amounts of penetration being noted and recorded as shown in table I. The concentrations of the dyes used were from 0.1 per cent. down to 0.001 per cent. In the case of the basic dyes, the higher concentrations caused more rapid infiltration of the tissue. In the case of the acid dyes, the lower concentrations frequently showed little or no staining at the apices and bases of the hypocotyls. The results of one experiment are summarized in table I, sections showing indistinguishable staining or infiltration being discarded. Three other experiments, run at other times, showed essentially the same thing, so that the table represents a typical case, and a confirmation of WENT's experiments. The numerals are represented on the same scale as in WENT's tables for purposes of comparison.

For purposes of comparison, WENT's averages for negative dyes were: apex 2.5, base 1.1; for positive dyes, apex 2.4, base 3.7. It is noteworthy that the greatest penetration WENT obtained was, at the most, one millimeter; while as much as three millimeters penetration was observed in the present work. Presumably sucrose in the solutions maintained oxidations and kept the tissues in a more normal condition during the time they were immersed.

RAMSHORN (**68**) criticized WENT's conclusions from such dye experiments, on the ground that actual electrical measurements of *Impatiens* cuttings showed electropositivity of the cut apices with respect to the cut bases. Upon repeating such measurements, measuring the dye uptake at the same time, quite the opposite was found. The experiment below is typical:

> *Impatiens* hypocotyls were cut and placed horizontally with each cut end in a cup of the dye solution in SHIVE's solution made up to 1 per cent. in sucrose. Agar–0.1 N KCl bridges from each cup led to a 0.1 N KCl solution in a cup in which the side-arm of a Zn-saturated $ZnSO_4$ half-cell could be placed. The potential differences between apex and base were measured with the WULF string electrometer described later in this paper. Table II shows the potential differences (P.D.'s) expressed in millivolts (mv.), the polarity being expressed as the sign of the tip with respect to the base. The dye penetration is represented in millimeters, the recorded figures being the averages. Several (3 to 5) sections were used for each dye solution, hence the electrical measurements represent the average of the several cuttings in parallel circuit. The dye concentrations were 0.05 per cent. All dye-charges were rechecked electrophoretically. The cuttings varied from 6 to 10 mm. in length.

In other experiments, the P.D.'s were read frequently over a period of several hours. It was revealed that the apex is at first positive[2] with respect to the base. In two hours after setting up the experiment it is electronegative, and remains so until the tissues appear abnormal (flaccid), after which the P.D. drops toward zero. This time-relation in establishing electrical polarities will be discussed in more detail under the section on P.D. gradients.

These electrical measurements confirm the dye-uptake experiments, and thus the polarity first claimed by WENT is real; that is, the apex of the *Impatiens* hypocotyl is electronegative to the base.[3]

RAMSHORN'S conflicting results may be explained by the time-relations in establishing the normal electrical polarity. It will be noticed that the apical negativity did not appear at once, but two hours or more elapsed before the tip became negative.

TABLE II

MEASURED ELECTRICAL POLARITY AND BIPOLAR DYE UPTAKE IN *IMPATIENS* HYPOCOTYLS*

DYES USED	POTENTIALS		DYE UPTAKE	
	6:00 P.M.	12 HR. LATER	APEX	BASE
	mv.	*mv.*	*mm.*	*mm.*
Negative dyes				
Light-green	+11.0 (tip +)	−14.0 (tip −)	1.5	0.0
Methyl blue	+13.0	− 5.0	trace	trace
Methyl orange	+12.0	− 7.0	7.0	5.0
Methyl orange-G	+15.0	− 5.0	1.5	0.0
Average	+12.7	− 7.7	2.4	1.2
Positive dyes				
Safranin	+ 9.0	− 7.0	trace	1.5
Methyl violet	+ 7.0	−12.0	1.0	2.0
Bismark brown	+ 3.0	− 3.5	trace	trace
Methylene blue	+ 1.0	− 1.5	0.5	1.5
Cresyl violet	+ 6.5	− 2.0	1.0	2.0
Brilliant cresyl blue	+ 9.0	− 9.0	trace	trace
Nile blue A	+ 6.0	− 9.5	1.2	2.5
Auramine	+ 9.0	− 6.0	5.0	7.0
Average	+ 6.3	− 6.3	1.1	2.1
Controls				
Shive's solution	+ 8	−12	.....	.....
Crone's solution	+ 7	−20	.....	.....

* The hypocotyls were cut and placed in the cups at 5:00 P.M. (3 per dye).

[2] In this paper the electrical polarity is expressed with respect to the external circuit.

[3] The possibility remains that the electrical polarity revealed by dye uptake and the polarity revealed by measurements are alike by coincidence.

2. MEASURED ELECTRICAL POLARITY.—

a. *Brief review of the literature.*—Many attempts have been made to correlate morphological and physiological polarity with electrical polarity in living organisms (*cf.* RAMSHORN, **68**; and WENT, **82**). CHILD'S school (CHILD and HYMAN, **17**; HYMAN and BELLAMY, **33**) claimed that the regions of highest metabolic rate in hydroids (apical regions) may be electronegative to other regions: LUND (**47, 50**), and LUND and KENYON (**53**), on the other hand, claimed that electrical polarity was dependent upon oxidation-reduction potentials (*cf.* discussion). Usually parts of polar tissues (apical end of hydroid stems, onion root tips) having the highest rates of oxidations were electropositive to other regions. RAMSHORN (**68**) stated that in seedlings of several different plants, electropositivity was directly linked with growth rate, such that potential distributions paralleled growth rate distributions. BARTH (**3**) showed that in several different hydroids the electrical polarity varied, some hydroids exhibiting apical electronegativity while others showed positivity. It is difficult to make *generalities* from such conflicting statements.

In general, the cortex of a root apex is held to be normally electropositive to that of the base (LUND and KENYON, **53**; MARSH, **55, 56**; RAMSHORN, **68**). In hypocotyls and coleoptiles of seedlings, the apical cortex is said to be electropositive to the basal (RAMSHORN, **68**). In the Douglas fir, the apical cortex is as RAMSHORN claimed to be the case in seedlings, *i.e.*, electropositive with respect to basal; while the apical wood is electronegative to the basal (LUND, **48, 49, 51**). In seedlings with internodes, in general, the nodal regions are electropositive to internodal zones; and the total polarity from apex (just below cotyledons) to the base of the stem shows an electronegativity of the tip to base (REHM, **70**; CLARK, this paper.) RAMSHORN (**68**), on the other hand, claimed the opposite, *i.e.*, that the tip was electropositive to the base (*cf.* discussion).

The following descriptions concern themselves directly with the determination of the electrical polarity of the *Avena* seedling, and of a few other seedlings.[4]

b. *Methods involved.*—In determining the electric potential differences (P.D.'s) in *Avena,* various types of electrodes, contacts, and recording instruments were tried. A Dolezalek electrometer was used as the recording instrument at first, but was found to be difficult of manipulation and to have too long a period. A Compton electrometer is subject to the same criticism, although it is somewhat better than the Dolezalek instrument. Lindemann electrometers are too insensitive. A potentiometer is apt to draw some

[4] Seedlings are preferred to mature, green plants because they have been more extensively studied, are more quickly obtained, and because they can be used in the dark. Light complicates the physiological behavior.

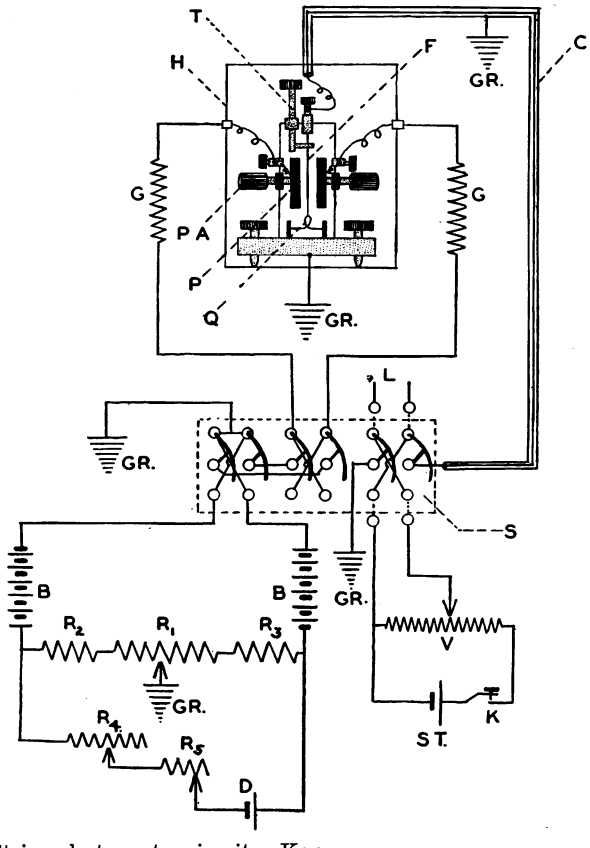

FIG. 1. String electrometer circuit. Key:

B	Six 45-volt heavy-duty Burgess B-batteries (long shelf-life)
C	Copper-tube shielding for fiber lead
D	1.5-volt dry cell battery
F	Platinum fiber (0.001 mm. diam.)
G	750,000-ohm wire-wound grid leaks
GR	Ground
H	Constant temperature housing
K	Key
L	Leads to unknown P.D.
P	Plates
P.A.	Plate adjustments
Q	Quartz fiber spring
R_1	10,000-ohm Yaxley wire-wound potentiometer
R_2, R_3	25,000-ohm Yaxley wire-wound potentiometers
R_4	750-ohm Yaxley wire-wound potentiometer
R_5	50-ohm Yaxley wire-wound potentiometer
S	Ceresin-covered mercury-in-paraffin switches
ST	Eppley standard cell
T	Tension adjustment
V	0.1–10,000-ohm plug-type Welch volt box

current from the living tissues. Hence a WULF string electrometer, constructed in the shops of this institute, was employed for the majority of the measurements (*cf.* WULF, **84**). Figure 1 represents the hook-up.

The sensitivity of the instrument in its final adjustment was more than one millimeter scale-deflection per millivolt with a scale-distance of one meter. Its period was about one second at lower sensitivities. The sensitivity depended upon the diameter of the string (platinum fiber 0.001 mm. in diameter), its tension, the distance between the plates, and the voltage across the plates. At higher sensitivity, damping increased the period to about three seconds. (This could be avoided by housing the instrument in a vacuum). The calibration curves approximated a straight line and remained constant for weeks at a time when the instrument was kept dry and at constant temperature. Because of the constant calibration and rapidity of motion of the string, readings could be made rapidly and accurately.

The distribution of potentials in the *Avena* seedling was first studied by moving contacts up and down the plant. CHAMBERS'S micromanipulators (40-pitch threading) were employed to move the electrode contacts.

Electrodes of various types were tried. It was found that bright or platinized platinum electrodes, however cleaned, gave non-reproducible readings, presumably because they were easily unpoised (*cf.* GICKLHORN, **26**; UMRATH, **77**; DORFMAN, **24**). Ag–AgCl wire loops or claws serving as contacts gave reproducible readings for a while, but demanded frequent replating. Quartz capillaries filled with fresh coagulated egg-white into which Ag–AgCl wire electrodes were set, gave reproducible readings. These were used for obtaining the internal distribution of P.D.'s. The gradients obtained with the Ag–AgCl loops and the quartz micro-electrodes, in general, gave similar results, these results being statistically comparable with those obtained with the more refined glass contacts and unpolarizable electrodes, although there was less constancy. The electrodes finally used for most of the measurements were Zn-saturated $ZnSO_4$ half-cells (*cf.* fig. 2). These remained iso-electric for months at a time. The types of contacts used in conjunction with these electrodes were varied. Usually the electrode was placed in a glass cup filled with the liquid used to make contact with the plant. A glass side-arm connected this electrode cup to the plant. At the point of contact, the side arm was fashioned into a small glass claw or loop, through which the plant led. The glass parts did not usually touch the plant, a meniscus of contact fluid performing this function. Tap water, KCl solutions, distilled water, and various nutrient solutions used in the glass contacts made no difference in the values obtained, if the same fluid was used in all contacts (*cf.* REHM, **69**). AMLONG (**1**) showed that geo-electric P.D.'s in plants depend upon the concentration of the contact fluids.

The effects of concentration were not determined in the present work, but it was assumed that the P.D's measured were not a function of the ionic species of the solution, since any of the solutions gave the same polarity, and approximately the same magnitude of P.D. (*cf.* REHM). The type of contact used for such measurements is seen in figure 2.

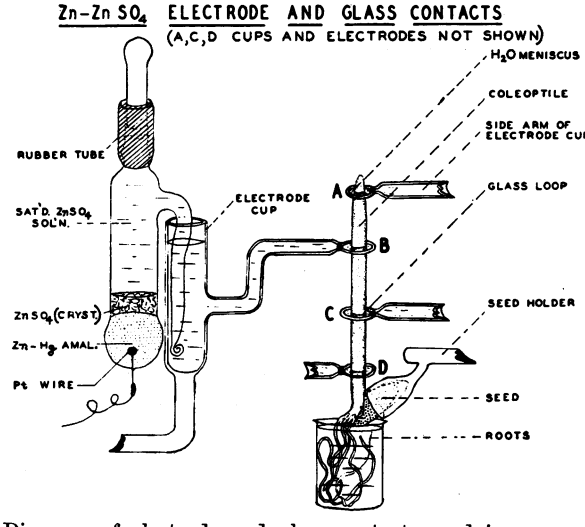

FIG. 2. Diagram of electrode and glass contacts used in measuring the P.D. of *Avena* coleoptiles.

A variation of this type of contact consisted in the use of a cotton or linen thread which was made wet by being threaded through the side arm of the electrode cup to the fluid in the cup. This thread then could either be wrapped around the part of the plant being studied, or could merely touch it (REHM, **69, 70**). Such threads maintained an isoelectric condition satisfactorily if they were occasionally washed.

The most useful type of contact, shown in figure 3, consisted in the following: Long, thin agar threads (made up of 2 per cent. agar in tap water, distilled water, 0.1 N KCl, SHIVE'S, CRONE'S, or HOAGLAND'S solution), 0.5 mm. or less in diameter, were made to hang from paraffined glass capillaries filled with the same agar. This was done by pushing an agar-filled capillary into more of the same agar, thus partly displacing the agar in the capillary as a thread. These capillaries were set in larger paraffined glass tubes filled with the same agar, which in turn were mounted in upright rows in a moist chamber. The glass tubes filled with agar led outside the chamber to paraffined electrode cups filled with the solution of which the agar was made. $Zn–ZnSO_4$ electrodes were placed in these cups. The agar threads hanging from the capillaries made contact to seedlings in the moist chamber,

being held to the plant by a drop of 15 per cent. gelatin. Several such fixed contacts could be made to each plant; and several plants could be set up simultaneously. The threads were prevented from drying by maintaining the chamber at near saturation with water vapor from strips of moist filter paper on the sides of the chamber. This method has the advantage that the agar threads remain fixed to the plant in the same position, being carried by upward growth without stimulating the plant. Seedlings, to which several glass contacts were fixed, frequently grew up through the more apical contacts, necessitating moving the apical contact back up to the tip. Such manipulations, however carefully done, usually stimulated the plant, thereby altering the P.D.'s.

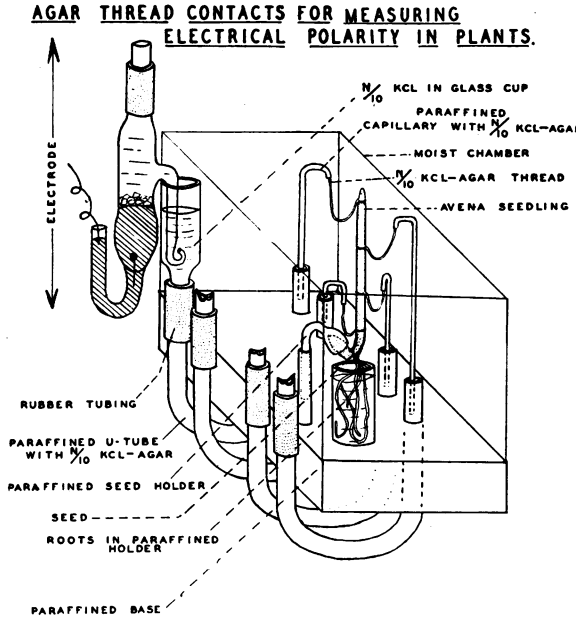

FIG. 3. Diagram of agar-thread contacts for measuring electrical polarity. Note that only one of several plants is shown, and only one of four electrodes is shown in the drawing.

c. *Electrical polarity and P.D. distributions in the Avena coleoptile.*— As described in an earlier paper (CLARK, **20**), the P.D.'s in the *Avena* seedling, obtained by manipulation of two contacts up and down the plant, were not constant over any considerable period of time, since the manipulation always resulted in changes of P.D.'s. Such "handling reactions" are illustrated in figure 4. This was usually true regardless of the care taken in making the manipulation, even if contact to the plant was made merely by a meniscus of water from the contacts. Moreover the orange light in the

dark room proved to be a stimulus.[5] Plants left in complete darkness gave variable P.D.'s as soon as this light was again turned on. Again, if two contacts were left on the plant in a fixed position, one at the tip and the other at the base of the coleoptile, and the plant left in complete darkness, con-

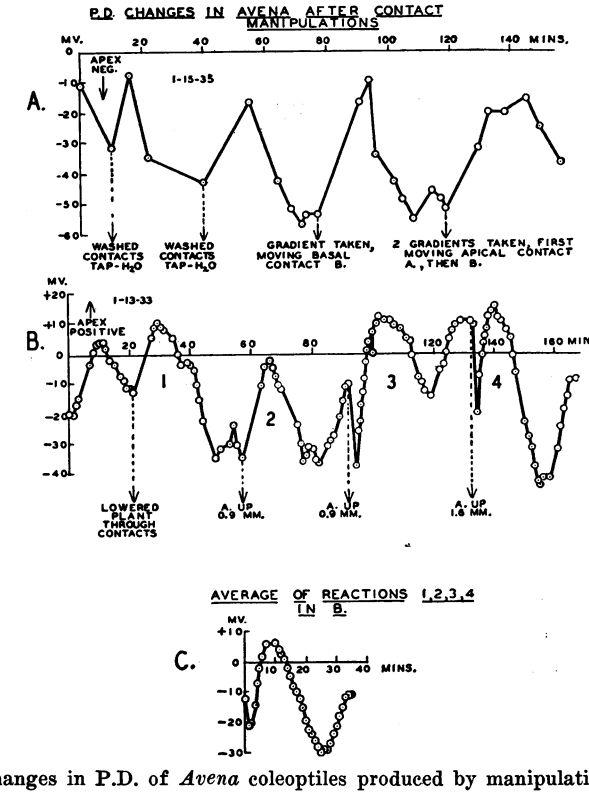

FIG. 4. Changes in P.D. of *Avena* coleoptiles produced by manipulation of contacts.

stancy of the P.D. was established only after an average time of 110 minutes. This constancy was abolished occasionally by the appearance of rhythmical P.D. changes, due probably to growth of the coleoptile up through the contacts, and also due to nutations which cause rubbing against the glass of the contacts. Clean glass adheres rather strongly to the *Avena* coleoptile cuticle, and considerable stimulation is caused by such movements. This rhythmical effect was avoided by the use of glass contacts dipped into 15 per cent. gelatin, thereby rendering the contacts slippery to the cuticle, or by recourse to the agar thread technique described above.[6]

[5] The light used to illuminate the dark room was filtered through a Corning filter no. 348, which cut out all wave-lengths below 575 mμ. No phototropism occurred in this light.

[6] Stimulation by contact is discussed by PFEFFER (65).

To obtain the normal P.D. distribution in the coleoptile, therefore, four fixed contacts were made to the plant (*cf.* fig. 2, A, B, C, D, and fig. 3). The plants were left in complete darkness. After constancy of P.D.'s obtained (80–120 minutes), the P.D. distribution could be easily and quickly determined. Figure 5 A represents the relative constancy of the P.D. between tip and base of *Avena* coleoptiles with fixed, gelatin-dipped glass electrodes; while figure 5 B represents the same type of experiment in which, however, the contacts were not gelatin-dipped.

Figure 5 C shows the much greater constancy obtained when the agar-thread contact method was used. The constancy lasts several hours. The figure has fewer points than either 5 A or B, but that no changes occur between the points has been verified by many other determinations.

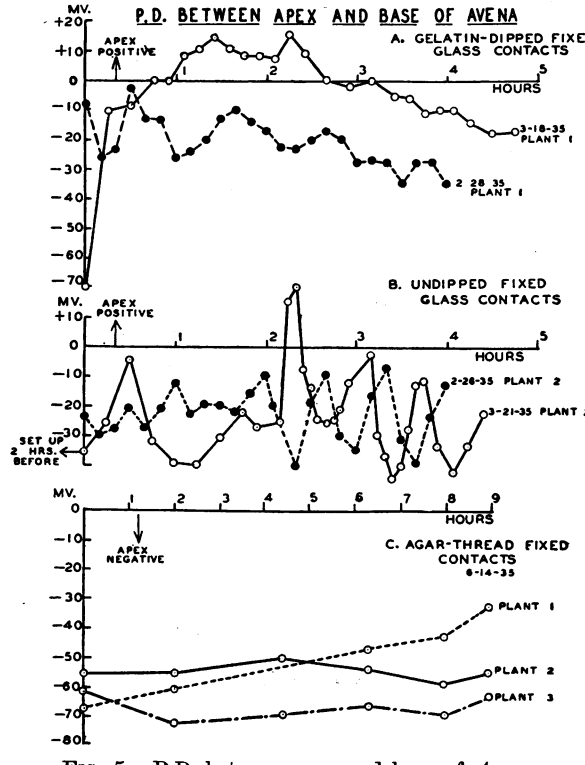

FIG. 5. P.D. between apex and base of *Avena*.

A. P.D. between apex and base of the *Avena* coleoptile, on using glass contacts dipped in gelatin.

B. P.D. between apex and base of *Avena* coleoptile, on using contacts not gelatin dipped.

C. Establishment of constant P.D. between apex and base of *Avena* coleoptile using agar-string contacts.

Figure 6 A illustrates the electrical polarity and P.D. distribution in the *Avena* coleoptile of intact plants as determined with the four fixed, gelatin-dipped glass contacts, the plant being in total darkness, and constancy having been obtained. The electrode cups each contained a $Zn–ZnSO_4$ electrode, each of which was isoelectric to all the others both before and after each experiment. The P.D. between each contact position to the plant was obtained by manipulation of mercury-in-paraffin switches outside the experimental chamber in which the plants were housed. Figure 6 B illustrates a similar gradient taken with only two glass contacts, the basal contact being fixed, and the apical contact being moved toward the base by means of the micro-manipulators. Every few mm., a reading was taken. After the contacts touched each other, the apical contact was again moved upward, readings being taken every 5 mm. The movements of such fluid contacts up and down the coleoptile did not wet the surface and thus invite electrical shunting (*cf.* ROSENE, **71**), because of the fatty nature of the cuticle. It will be noticed that the gradient taken by the manipulation down the coleoptile differs from that taken on moving the contact back up the coleoptile. After such manipulations it is found that the P.D.'s vary considerably, and sometimes the polarity is reversed for considerable time (*cf.* figure 4). Figure

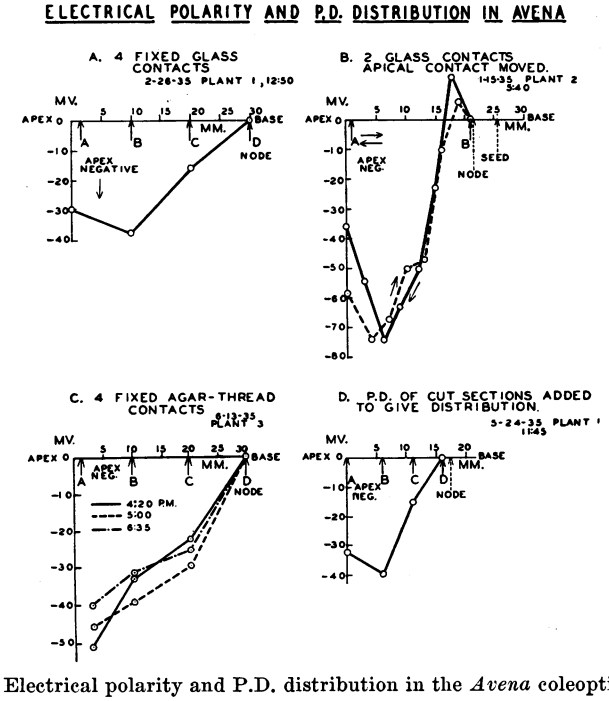

FIG. 6. Electrical polarity and P.D. distribution in the *Avena* coleoptile with various contacts and arrangements.

6 C represents a similar gradient, the contacts in this case being four fixed agar threads as described above. The three curves in C represent three gradients taken on the same plant at different times, showing that the P.D. distribution remained constant for a considerable time. Figure 6 D represents the P.D. distribution calculated from the individual P.D.'s of cut-sections of a coleoptile (*cf.* section on P.D.'s of cut sections). In all of these curves, electronegativity of the tip is represented on the ordinates, and the length of the coleoptile is represented on the abscissae, A, B, C, and D representing points of contact to the coleoptile from tip to base. The potential at D is taken as the reference zero. The P.D. from A to D is always equal to the sum AB + BC + CD.

From the above experiments it is clear that the tip of the *Avena* coleoptile is normally electronegative to the base.

d. *Electrical polarity and P.D. distribution in Pisum, Impatiens, and Zea.*—*Pisum sativum* seedlings were studied for the normal electrical polarity and P.D. distribution. Contacts were made to etiolated plants with the linen threads previously described, the threads being wrapped around the zone to be measured. There were several electrode contacts to each plant, represented by the positions on the abscissae corresponding to the points plotted in figure 7 A, 1, 2, 3. The P.D. obtained between each contact was found to be fairly constant for several hours at a time. The measurements were made in the darkroom in weak red light. Figure 7 A represents typical gradients. It will be noticed that nodal zones are electropositive to the internodal zones, and that there exists an electronegativity of the tip with respect to more basal regions. This confirms REHM'S (**70**) findings on *Phaseolus,* and disagrees with RAMSHORN'S (**68**) finding that the tips of *Asparagus* seedlings are electropositive to the basal regions, although the nodal zones were positive to internodal zones.

Etiolated *Impatiens balsamina* seedlings were set up in the same way as *Pisum* seedlings were, and the P.D. distribution recorded. The hypocotyls are without nodes, and the plants were only seven centimeters in height. Figure 7 B represents the distribution found.

Zea mays seedlings 5 to 7 cm. in height were set up in the same way as has been described for the *Avena* seedlings, four fixed contacts being made by means of agar threads. The polarity and P.D. distributions found corresponded very closely with those recorded for *Avena.*

It is concluded from these observations that in *Pisum, Impatiens,* and *Zea,* the tip of the etiolated seedling is normally electronegative to the basal regions. This shows that the electrical polarity of the *Avena* coleoptile is not unique in its apical negativity.

e. *Internal electrical polarity in the Avena coleoptile.*—The above discussion concerns the electrical polarity and P.D. distribution measured on

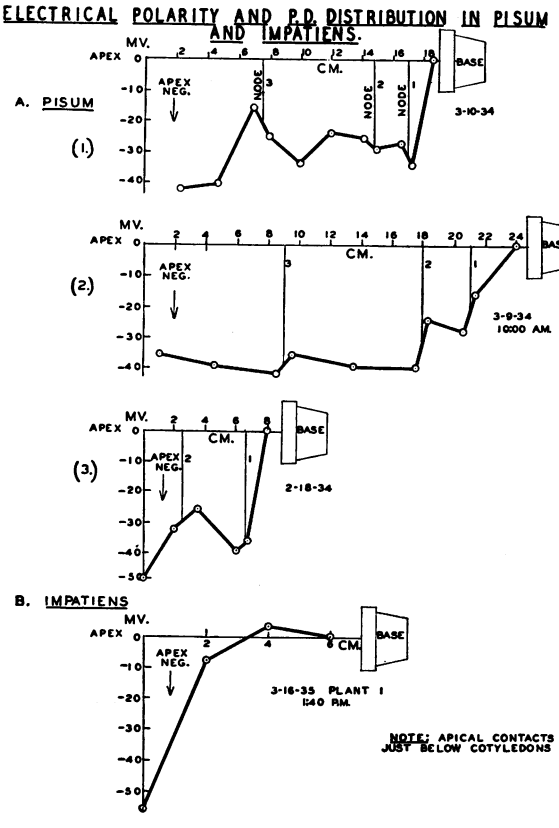

FIG. 7. Electrical polarity and P.D. distribution in *Pisum sativum* and *Impatiens balsamina.*

the cuticle, thus the *external* polarity. It is conceivable that the distribution of *internal* P.D.'s might be different, as suggested by LUND'S (**49, 51**) findings in the Douglas fir. Here the apical wood was electronegative to basal wood, while the apical cortex was electropositive to basal. For this reason a few experiments were performed on the *Avena* coleoptile, in which the P.D. distribution beneath the cuticle was examined. Quartz microelectrodes (*cf.* descriptions in this paper) were inserted in coleoptiles by means of micromanipulators, and the electrical polarity was measured. The electrodes are illustrated in figure 8. They maintained an isoelectric condition very satisfactorily for two or three hours. Figure 8 A shows the P.D. between two such electrodes inserted in a coleoptile. One electrode was inserted in the wall of the coleoptile at the apex, and one at the base, both electrodes being in the same side-wall, but being inserted from opposite sides of the coleoptile. Traumatropisms toward the sites of insertion occurred

after about 30 minutes, as diagrammed in figure 8 A. Immediately after insertion, the apex of the coleoptile became electropositive to the base, but after 20 or 30 minutes, as seen in figure 8 A, the tip became negative. It was considered impracticable to insert several such electrodes, or to reinsert the same two electrodes at several different loci on the coleoptile in order to obtain the P.D. distribution. This would have resulted in even more P.D. variation than is depicted in the figure; hence, in order to obtain the distribution of internal P.D.'s, a different technique was employed. Several longitudinal slits were made down the coleoptile using a sharp razor. Each slit had its counterpart on the opposite side of the coleoptile in order to compensate the wounding effects. Such coleoptiles will remain straight, whereas the ones in which the microelectrodes were inserted showed traumatropisms. Glass-loop contacts, such as were described above, made contact with the plant, one at the tip and one at the base of the coleoptile. The basal contact was racked up the coleoptile toward the apex by means of the micromanipulator. When a contact was centered over a slit, presumably the potential internal to the cuticle was measured. Figure 8 C shows the distributions obtained in this way. The vertical lines, a, b, c, d, and e represent the loci of the slits in the coleoptile. Curve I represents the distribution obtained by racking the basal contact up to the apex, whereas curve 2 represents the distribution obtained by racking it back down a few minutes later. Curve 3 represents the distribution obtained by racking it back up 20 minutes later. Figure 8 B illustrates the changes in P.D. between the apex and base of the coleoptile before and after each distribution was determined. It will be noticed in curve 2 of figure 8 C, that the readings were taken with the contacts centered on the slits. The distribution is very similar to that obtained on the intact cuticle. Curve 1 shows the same thing, with the exception that one contact was centered *between* two slits, thus on the intact cuticle (between d and e). Curve 3 shows the type of curve obtained when no attention was paid to the position of contacts with respect to slits, *i.e.*, the distribution was taken at more frequent loci, regardless of the slit positions. The general electrical polarity is the same as that on the intact cuticle, but the curves are not smooth. This indicates a *radial* P.D. between cuticle and the internal tissues. The curves are smooth if contacts and slits coincide.

The following section (f) will show that cut sections are of the same electrical polarity as has just been described; and, as seen in figure 6, the sum of the section P.D.'s of a coleoptile give a P.D. distribution for the coleoptile which is comparable to that in an intact plant.

The evidence presented indicates that the distribution of internal P.D.'s in the *Avena* coleoptile is the same as that on the outside (cuticle). The polarity is the same, *i.e.*, the tip is electronegative to the base.

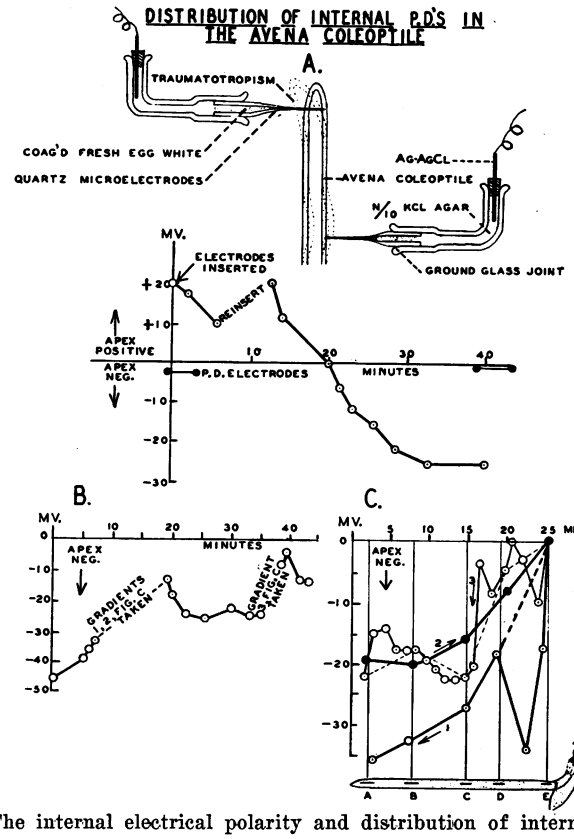

FIG. 8. The internal electrical polarity and distribution of internal P.D.'s in the *Avena* coleoptile.

f. *Electrical polarity of cut sections.*—The P.D.'s and electrical polarity of cut sections of the *Avena* coleoptile were then determined in several different ways. One method involved cutting the sections with two parallel razor blades separated by a brass strip. The cut surfaces were washed by placing the sections upright on wet filter paper for an hour. The sections were then carefully transferred to the experimental chamber, using eye-forceps. Contact was made to individual sections with the agar-thread method, or with agar strips. 0.1 N KCl agar was used in most cases. The strips or threads led through paraffined glass tubes to electrode cups outside the chamber. $Zn–ZnSO_4$ electrodes were placed in these cups, and the P.D.'s measured. The chamber was maintained at a high vapor pressure by means of strips of moist filter paper.

Several hundred measurements on 3-mm. sections revealed that the cut apical surface was always electronegative to the cut basal surface from 1 to 15 mv., the magnitudes depending upon the time at which the measurements were made.

Another method involved placing from 12 to 20 sections on an agar block, and placing a similar agar block on the tops of the sections. 0.1 N KCl agar strips made contact with these blocks and to the cups outside the chamber. Here, therefore, the average P.D. of several sections in parallel was measured. The same result was obtained, *i.e.*, apical negativity. This latter method will be discussed again in a later paper.

A third method involved making contact with several places on longer sections with agar threads held in place with a drop of gelatin, as described in an earlier section of this paper. The cut surfaces were usually electronegative to the intact cuticle (another indication of a "radial" polarity), but the apical cut surface was always negative to the basal cut surface.

Using the method by which 20 sections were measured in parallel at the same time, the relation of the length of the section to the P.D. of the section was determined. It was usually found that time was required before the maximum P.D. was established (Presumably this was due to the diffusion of ions from the sections into the agar blocks). For this reason, the P.D.'s were allowed to reach their maximum values before plotting against length. This time function was more pronounced in longer sections. Figure 9

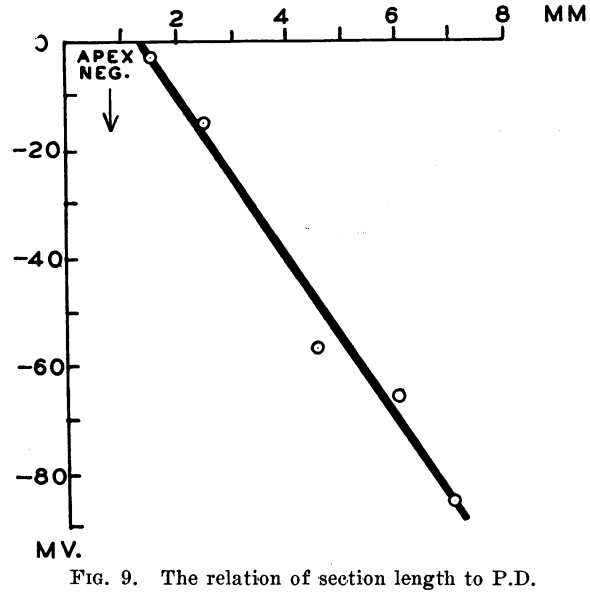

FIG. 9. The relation of section length to P.D.

shows the length of the section in millimeters plotted against P.D. in millivolts, after this maximum P.D. had been reached in all sections (5 hours).

In the section on P.D. distribution in intact plants (fig. 6), it was seen that the sum of the several P.D.'s along a coleoptile was equal to that measured from apical contact to basal contact. In figure 9 it is seen that

this principle of summation again holds, since the magnitude of the P.D. of cut sections is directly proportional to the length of the section. (*Cf.* LUND, **47, 51,** and ROSENE, **71,** on the principle of summation of P.D.'s.)

Cut sections of *Vicia faba* and of *Pisum sativum* showed the same electrical polarity as *Avena* coleoptile sections. In the section on dye-uptake, it was seen that *Impatiens* cuttings also showed apical negativity. Thus the phenomenon seems quite general. In *Vicia* and *Pisum,* the P.D. magnitudes varied from a few millivolts to 30 or 40 millivolts, depending upon the length of the section.

g. *Geoelectric effect.*—During the measurements of section P.D.'s, it was noticed that inverted sections exhibited an inverted electrical polarity. A section seemed to show negativity of the end oriented upward regardless of whether this end was morphological tip or base.

The experimental procedure usually consisted in inverting single sections and measuring the individual P.D.'s of these sections, or by placing 12 to 20 sections on one agar block, making contact with this block and with the other cut surface with a similar agar block, thus obtaining the average P.D.'s of the lot of sections in parallel. Measurements were made immediately, when possible, upon inverting the sections. The polarity of the inverted sections showed an immediate inversion of electrical polarity. The time relations of the inversion have not been carefully studied, but the establishment of the inverted polarity seemed to take less time than the geoelectric effects of BRAUNER (**13**). It was noticed, however, that this geoelectric effect was not maintained indefinitely, particularly in the shorter sections. The original polarity (apical negativity) returned within 60 to 120 minutes, depending upon the length of the sections. Figure 10 A shows the course of the P.D.'s of inverted sections during a period of time. The sections were cut at 9:30 A.M. and placed on wet filter paper in an inverted position. The experiment was set up at 11:30 A.M., the first readings being taken at 11:40 A.M. During this time, the inverted polarity had attained a considerable magnitude.

Figure 10 A, curve 1, shows the change in P.D. of control upright 3-mm. sections. They exhibit normal apical negativity. Curve 2 is for inverted sections 3 mm. in length; curve 3, 5 mm.; curve 4, 7 mm. in length. It is seen that the longer the sections, the greater the magnitude of the inverted polarity; and that the greater the magnitude of this inverted polarity, the longer the time necessary to reestablish the normal polarity. The abscissae are in hours after cutting the sections. Figure 10 B represents data from the same experiment. The P.D.'s of sections with inverted polarity are plotted against the length of the sections in millimeters. The P.D. values used are those of maximum magnitude, *i.e.,* at the first measurements, as represented on the zero ordinate of figure 10 A. It is seen that a nearly

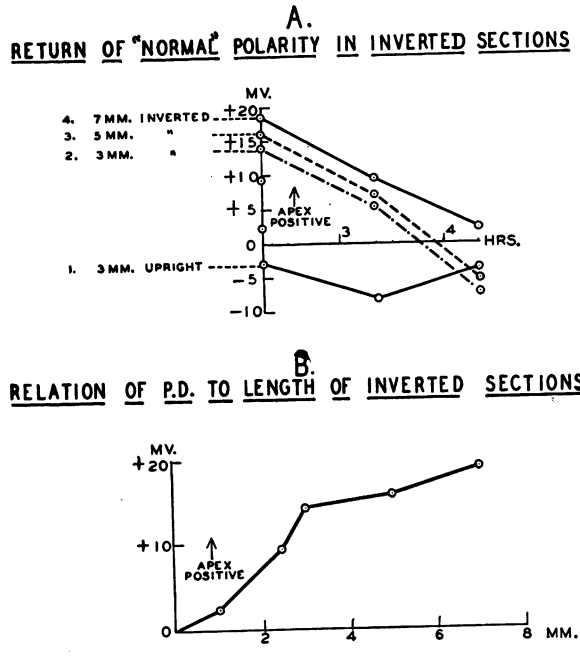

FIG. 10. "Geoelectric" polarity of sections.

direct proportionality exists between length and the P.D.'s of inverted polarity, thus indicating a similarity to upright sections which exhibit the same direct proportionality.

The possibility remains that the normal polarity of upright sections of plants is partly a result of the position of the plant or section with respect to gravity, *i.e.*, that the electrical polarity *is*, partly, a geoelectric effect. This scheme is complicated by the return to normal polarity with time. It is not known what the effect of inverting intact plants, *e.g.*, roots or shoots, has on their electrical polarity. This problem is being investigated. The establishment of positivity of the under side of inverted sections recalls to mind the similar establishment of positivity of the under side of horizontally-placed plants (BRAUNER **13**).

That the geoelectric polarity or inverted polarity is not a phenomenon confined to *Avena* sections was shown by the fact that 5-mm. inverted *Pisum* sections developed the inverted polarity. In this case, however, more time was required to establish the inversion. On first inverting, the polarity was found to be inverted a few millivolts, the maximum inverted polarity becoming established only after 3 or 4 hours. Since BRAUNER (**14**) showed that the geoelectric effect varied in the seed coat of various plants, depending upon the membrane structure, this is not surprising. It is likely that other

plant sections would exhibit their own peculiarities. This individuality was also observed by BRAUNER (**13**) for geoelectric effects in horizontally placed plants.

From the section f on electrical polarity, it was concluded that the normal, inherent electrical polarity of intact plants and cut sections of *Avena, Zea, Pisum,* and *Impatiens* was an electronegativity of the apical parts with respect to more basal parts. This polarity exists internally as well as externally, and is not directly related to growth.

Discussion and conclusions

It has been experimentally demonstrated that the normal electrical polarity of several seedlings is an electronegativity of tip to base. Several views are held as to the mechanism of the origin of this electrical polarity.

LUND (**47**) has claimed that electrical polarity is the result of oxidation-reduction potentials such that usually, but not necessarily, regions of highest rates of oxidation are electropositive to other regions (or, in the thermodynamic sense of red-ox potentials, the ratios of oxidant to reductant are different in the different ends of the structure concerned). A detailed discussion of the theory is out of place at this time (LUND, **50**), but certainly the theory is thrown into doubt by the fact that red-ox potentials can be measured only by indifferent electrodes, and not by non-polarizable electrodes such as used by LUND. FRANCIS (**25**), BEUTNER AND LOZNER (**5**), RAMSHORN (**68**), STERN (**76**), and DORFMAN (**23, 24**), have all criticized LUND's theory on this basis. STERN (**76**) and MARSH (**56**) stated that if the living membranes, to which LUND made contact with non-polarizable electrodes, acted as indifferent, metallic conducting electrodes, the P.D. measured could be the same as LUND's hypothetical oxidation-reduction chain interposed between the contacts. While it is improbable that these membranes act as metallic conductors, yet the possibility remains that they may do so.[7]

Bioelectric potentials, as linked with oxidative processes, can be explained by other mechanisms as well as by oxidation-reduction potentials (STERN, **76**; DORFMAN, **23, 24**; BEUTNER AND LOZNER, **5**; FRANCIS, **25**). Most of these explanations are based on the effects of oxidations on diffusion potentials or membrane potentials.

The oxidation-reduction polarity theory, moreover, is thrown into doubt by the experiments of DORFMAN (**23, 24**), who showed that the oxidation-reduction polarity of the frog's egg was opposite in sign to the bioelectrical polarity measured with non-polarizable electrodes.

[7] SCHOTT and BORSOOK (72) have shown the possibility of metallic electron conduction between enzyme centers in *E. coli.* FETCHER (dissertation, University of Chicago, 1934) demonstrated the possibility of electron conduction in membranes which are composed of conductors of the second class (LILLIE, 44).

The bioelectrical polarity of the *Avena* coleoptile is certainly not directly linked with respiration in the different regions of the coleoptile, since BONNER (**6**) showed that there was no distribution of respiration in this organ. Moreover, from data to be published shortly, reduced ascorbic acid (vitamin C) is found in highest concentration in the apex of the *Avena* coleoptile, the concentration decreasing basally (cystine, cystein, and glutathione are not present). It was also indicated that the reverse relation held for the distribution of oxidized ascorbic acid. This does not conform with LUND'S oxidation-reduction polarity (LUND, **50**), since by this theory, the tip would usually be electropositive. This is assuming, however, that the oxidation-reduction potentials of ascorbic acid could play a part in the electrical polarity. (For exceptions to this polarity rule, see LUND, **50**).

With regard to the disagreement between the findings presented in this paper and those described by RAMSHORN (**68**), the following discussion becomes pertinent: RAMSHORN made measurements of the electrical polarity of several different seedlings and roots, and showed a parallelism between growth and electropositivity. Regions of highest growth-rate were electropositive to other regions. Temperature changed both in the same way, and applied potentials accelerated growth if the applied polarity coincided with the measured inherent polarity; and, conversely, inhibited growth if the polarities were opposed. On page 741 of his paper, RAMSHORN presents a series of curves of the gradient of electric potentials from tip to base in *Helianthus* hypocotyls after stimulation by shaking. After stimulation, the tip became electronegative to the base presumably within a few seconds. In 15 minutes the tip became electropositive, and in 75 minutes the magnitude of this positivity had diminished only a few millivolts. This roughly confirms LUND'S (**52**) finding that the electropositivity of the apex of the Douglas fir decreases or that the tip even becomes negative on mechanical stimulation.

In the present study of the *Avena* coleoptile, reliable constancy of P.D.'s was not obtained until 90 to 110 minutes after setting up the experiment, as much care as possible being taken not to stimulate the plants during this operation. Moreover, constancy was not good unless the plants were in complete darkness; and manipulation of contacts from point to point involved considerable stimulation.

RAMSHORN'S correlation between growth and electrical polarity might, in my opinion, suffer a reversal in some cases, particularly in *Avena,* if the time relations, contact manipulation, and light conditions of his experiments were reinvestigated, especially with regard to the constancy of observed P.D.'s over longer periods of time.

BARTH (**3, 4**) observed that apical positivity and organic polarity in the hydroids are not correlated, but that either apical or basal positivity

may be correlated with organic polarity, depending on the hydroid used. This lends no support to RAMSHORN'S positivity theory; nor do REHM'S (**70**) measurements on *Phaseolus,* in which he found the tip of the plant electronegative to the basal regions.

CZAJA (**21**) says that the auxin-transport itself electrically polarizes the plant, thus roughly supporting RAMSHORN'S statement that auxin changes the P.D.'s and growth rate. CZAJA, however, is largely theoretical in his consideration, and bases his assumptions on results obtained with unphysiological concentrations of auxin. In the light of unpublished experiments of my own, the effect of auxin on plant potentials is a real one, but possibly one not closely linked with the normal inherent polarity. DE HAAN (**30**) is also of this opinion.

Experiments on the effect of gravity on the *Avena* coleoptile P.D.'s have revealed that the electrical polarity can be changed or inverted by inverting their morphological axes. This polarity inversion is not permanent, the original polarity returning. Hence "normal" electrical polarity is not exclusively and directly caused by geoelectric potentials; but it is possible that they are contributing factors. This possibility is being examined.

The mechanism of the origin of electrical polarity might be linked with the phenomenon of polar conductance observed, *e.g.,* by BRAUNER (**14**), METZNER (**59**), and GUHA (**29**). Plant tissues have been shown to exhibit a selective ionic permeability so that an electric current is conducted more easily in one direction than in the other. Investigations are under way to see if there is any relation between polar conductance, electrical polarity, and polar transport of auxin.

Summary

1. Theories of the cause of polar transport of auxin in plants are discussed. The electrical theory has been accepted by many workers as one of the most plausible (*cf.* second paper, following issue of PLANT PHYSIOLOGY).

2. WENT'S bipolar dye-uptake experiments on *Impatiens* cuttings, used to substantiate his electrical transport theory, are repeated and confirmed. Positively charged dyes are taken up most by bases, negatively charged dyes by apices, of *Impatiens* cuttings. This is in agreement with the fact that electrical measurements show that *Impatiens* cuttings have apical electronegativity.

3. Intact *Avena* and *Zea* coleoptiles, *Pisum* stems, and *Impatiens* hypocotyls exhibit apical negativity when constancy of P.D. measurements is obtained. Various methods of measuring this polarity are discussed.

4. Cut sections of *Avena* and *Zea* coleoptiles, and of *Pisum* and *Vicia* stems exhibit the same polarity found in *Impatiens* cuttings, *i.e.,* apical negativity. Time is required to establish this polarity. The P.D.'s of sections are directly proportional to the length of the sections.

5. The internal electrical polarity of the *Avena* coleoptile is the same as the external.

6. Inverting sections inverts their electrical polarity, *i.e.*, the morphological apices become electropositive to the bases. This inverted polarity disappears with time. It is proportional to the length of the sections as in the case of upright sections; and the time of disappearance of the inverted polarity is proportional to the length of the sections.

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